

Fragmentation of colonial ascidians: Differences in reattachment capability among species

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When colonial invertebrates (e.g., sponges, corals, gorgonians, etc.) are damaged and torn into fragments, the resulting fragments often survive and can reattach to appropriate substrata (e.g., Tsurumi and Reiswig, 1997; Smith and Hughes, 1999; Schonberg and Wilkinson, 2001). Thus, fragmentation can act as a reproductive and dispersal mechanism for these animals. Biologists have long known that colonial ascidian fragments can reattach to the substratum and researchers have often used this property to their advantage during experiments (e.g., Bullard et al., 2004). Very few studies, however, have specifically described the reattachment capability of colonial ascidian fragments (Berrill, 1951; Stoner, 1989; Worcester, 1994; Edlund and Koehl, 1998) and none has assessed the probability of successful reattachment by fragments empirically. It remains unclear whether fragments of all colonial ascidian species can reattach successfully or if there are species-specific differences in reattachment ability.

We conducted our study in the eastern end of Long Island Sound (USA) at the University of Connecticut's Avery Point campus in Groton, Connecticut. Floating

docks at this site harbor a diverse fouling community including numerous colonial ascidians, many of which are invasive (see Osman and Whitlatch, 2004 for details on the site description). We conducted two sets of laboratory assays to assess the ability of fragments from colonial ascidians to reattach to the substrata. In the first assay, fragments of four species (*Aplidium constellatum*, *Botryllus schlosseri*, *Botrylloides violaceus*, and *Didemnum* sp. A) were placed on PVC panels (100 cm²) in a flow-through seawater table with unfiltered water for 30 h. Field-collected ascidian colonies were torn into similar-sized fragments (ca. 3 cm diameter) with forceps. Each fragment was then placed right-side up in the center of a panel in the water table. After 30 h fragments were squirted with a pipette to determine if they had attached to the panels; attached fragments remained in place, unattached fragments fell off the panels. To determine if attached fragments were strongly or weakly attached, panels with attached fragments were removed from the water table and held upside down for 2 s. Fragments that remained attached after 2 s were considered strongly attached while those that fell off were considered weakly attached. A single Chi-square analysis was used to determine if there were differences in the number of fragments attached after 30 h for the four species of ascidians. Three separate Chi-squares

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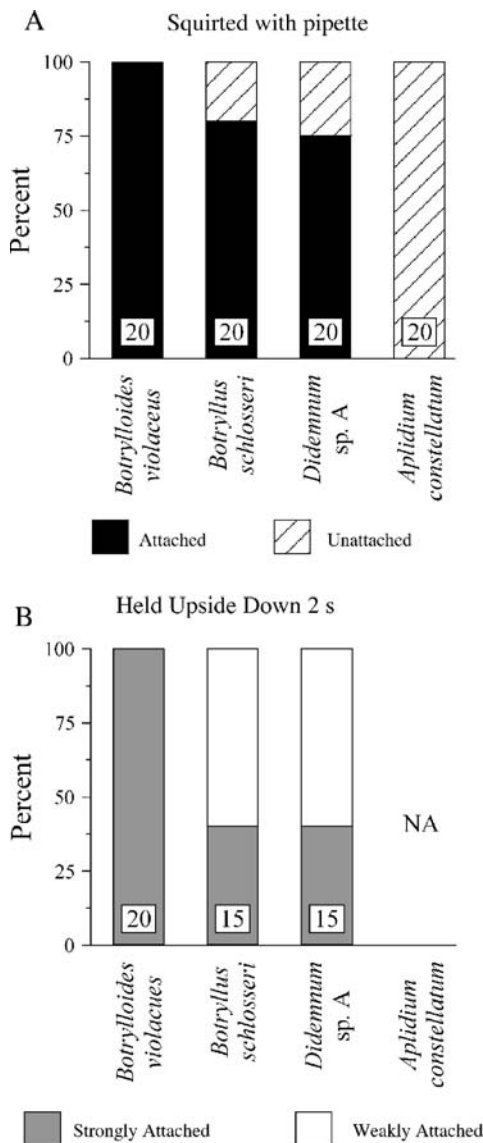


Fig. 1. Attachment of colonial ascidian fragments to PVC panels after 30 h. Sample size is indicated in the box at the base of each histogram. A. Percentage of fragments of different species attached (not blown off by a pipette) after 30 h. B. Percentage of fragments of different species strongly attached (able to be held upside down out of the water for 2 s) and weakly attached (could not be blown off the PVC with a pipette, but fell off when held upside down) after 30 h.

were used to compare differences in adhesion strength between species, one analysis for *Botrylloides violaceus* versus *Botryllus schlosseri*, one for *B. violaceus* versus *Didemnum* sp. A and one for *B. schlosseri* versus *Didemnum* sp. A.

For the second assay, fragments of *Botryllus schlosseri*, *Botrylloides violaceus* and *Didemnum* sp. A were placed on panels in a water table (as above) and

checked for reattachment every 6 h for 30 h. Fifty fragments of each species were used, each from a separate colony. Ten fragments of each species were randomly chosen every 6 h and squirted with a pipette to check for attachment to the PVC panels (as above).

Significant differences in the ability of fragments of different colonial ascidian species to attach to the substrata were found ($P < 0.001$; Chi-square) (Fig. 1A). Fragments of *Aplidium constellatum* did not reattach to PVC panels after 30 h, while most fragments of *Botrylloides violaceus* and *Didemnum* sp. A, and all fragments of *Botryllus schlosseri* reattached. Fragments of *B. violaceus* adhered more strongly to the substrata than fragments of *B. schlosseri* or *Didemnum* sp. A ($P < 0.001$; Chi-square in both cases) (Fig. 1B). All *B. violaceus* fragments remained attached to PVC panels when held upside down for 2 s while only 40% of *B. schlosseri* and *Didemnum* sp. A fragments remained attached. There was no difference in the adhesion strength between *B. schlosseri* and *Didemnum* sp. A ($P = 1.00$; Chi-square) (Fig. 1B). Fragments of *B. schlosseri*, *B. violaceus*, and *Didemnum* sp. A reattached to the substratum quickly; some fragments of all three species reattached in as little as 6 h while many reattached within 12 h (Fig. 2).

Clearly, there are species-specific differences in reattachment ability of colonial ascidian fragments. Fragments of some colonial ascidians readily reattach while others do not. In our assays, fragments of *Botrylloides violaceus* almost always successfully reattached while fragments of *Aplidium constellatum* never reattached. *Botryllus schlosseri* and *Didemnum* sp. A also had high levels of reattachment, with about 75–80% of fragments reattaching after 30 h. In the field, colonial ascidians could become fragmented by physical or biological disturbances (such as storms, accidental impacts of organisms with the bottom, predation,

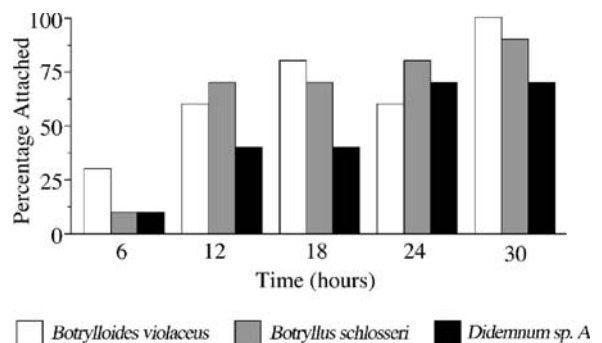


Fig. 2. Percentages of colonial ascidian fragments attached (not blown off by a pipette) to PVC panels over a 30 h period. Sample size equals ten separate fragments assessed for each species at each time period.

herbivore browsing, or other means). Once fragments are produced, they may be transported by currents, waves, and tides. Many fragments may be washed into unsuitable soft-sediment habitats and eventually die (e.g., Coutts, 2002). Other fragments may become lodged against rocks, eelgrass blades, docks, pilings, or other hard substrata. If they remain in contact with hard substrata long enough, fragments may reattach and thrive at their new location. Thus, fragmentation may provide a means for asexual reproduction and dispersal for some colonial ascidians.

Differences in reattachment ability could be related to the life-history ecology of different ascidian species. Some species, such as those that produce very thick and fleshy colonies (e.g., *Aplidium constellatum*), may only rarely become fragmented. Other species may commonly use a fragmentation strategy for asexual reproduction and dispersal. For example, *Didemnum* sp. A possesses fragile, lobe-like morphologies that readily produce fragments (Bullard et al., 2006-this volume). *Didemnum* sp. A is rapidly spreading along both coasts of the U.S. and fragmentation may allow it to quickly colonize new areas. Additional information needs to be collected to determine the natural rate of fragmentation in colonial ascidians, the dispersal rates of fragments and whether these vary between species and habitats.

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